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Original communication

The role of the Armanni-Ebstein lesion, hepatic steatosis, biochemical analysis and second generation anti-psychotic drugs in fatal diabetic ketoacidosis

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ABSTRACT

Diabetic ketoacidosis (DKA) is an acute severe complication of diabetes and characterized by a complex disordered metabolic state due to an absolute or relative insulin deficiency, leads to hyperglycaemia, ketoacidosis and ketonuria. DKA can cause sudden unexpected death and often yields minimal and/or subtle autopsy findings or a negative autopsy and the diagnosis mainly depends upon biochemical analysis of body fluids. This communication highlights the role of Armanni-Ebstein lesion, hepatic steatosis, biochemical analysis and second generation anti-psychotic drugs in 25 adult cases of fatal diabetic ketoacidosis. The study recognises and reconfirms that fatal DKA occurs in both type I and II diabetes. The macroscopic autopsy features observed in this study are non-specific and do not guide the pathologist towards the diagnosis of fatal DKA. Once other possibilities have been excluded, the Armanni-Ebstein lesion alone or the combination of hepatic steatosis and Armanni-Ebstein lesion in an otherwise negative autopsy of a sudden unexpected death should raise the suspicion of DKA as the cause of death and indicate biochemical analysis of body fluids. Our findings also remind forensic pathologists to search for fatal DKA in sudden unexpected death with a negative autopsy, where there is a history of second generation anti-psychotic treatment.

1. Introduction

Diabetic ketoacidosis (DKA) is an acute severe complication of type I diabetes mellitus (DM) and is not uncommon in type II DM. DKA, characterized by a complex disordered metabolic state due to an absolute or relative insulin deficiency, leads to hyperglycaemia, ketoacidosis and ketonuria. The mortality rate in patients with DKA is 7–8%.[1]

There are important medicolegal issues associated with DKA. DKA can cause sudden unexpected death and often yields minimal and/or subtle autopsy findings or a negative autopsy. Post-mortem diagnosis mainly depends upon biochemical analysis of body fluids. If decompositional changes are present, the value of biochemical analysis gradually decreases, and arriving at a diagnosis is further complicated. Vitreous fluid is better protected from decomposition than is blood, and therefore this has become the most popular specimen for analysis.[2] DKA can be identified by highly elevated levels of blood acetone[3] and the presence of a high glucose value accompanied by ketones in the vitreous humour.[2]

Armanni-Ebstein lesions are subnuclear vacuolations of the renal tubular cells which contain fat and were first described by Luciano Armanni in 1877 and by Ebstein in 1882.[4] These lesions strongly indicate death in diabetic coma.[5] In addition, Armanni-Ebstein lesions can be seen in non-diabetic ketoacidosis resulting from alcoholism,[6] starvation,[7] and hypothermia.[8] Hepatic steatosis, a non-specific finding seen in diabetes, occurs in a variety of other pathological conditions as well. Diabetic glomerulosclerosis is an indication of long-standing diabetes and increases with the duration of diabetes.[9] Hepatic steatosis and diabetic glomerulosclerosis do not substantiate DKA. Although the role of second generation anti-psychotic drugs causing DKA is well known to clinicians, it is under-recognized as a cause of DKA in sudden unexpected deaths with negative autopsy findings.

2. Methods

All adult autopsy cases in which the cause of death was given as DKA, during a period of five years (2005–2010) in the Provincial

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Forensic Pathology Unit, Toronto, Ontario, Canada, were selected. This unit has the jurisdiction for the City of Toronto. Cases with incomplete files and/or microscopic sections showing autolytic changes were excluded.

In each case, the completed post-mortem report, Coroner's Warrant and/or police occurrence report, toxicological report issued by the Centre for Forensic Sciences and histological slides were reviewed. When available, past medical records and vitreous fluid analysis reports were reviewed. The analysed data included clinical history, sex, age at death, terminal event, macroscopic findings at autopsy, hepatic steatosis, Armanni-Ebstein lesion, diabetic glomerulosclerosis, as well as blood acetone and vitreous glucose and ketone levels.

3. Results

Twenty-five adult cases comprised the study population (Table 1) including cases of both type I and type II diabetes. Except for one, all were found dead and not resuscitated. There were twenty males (80%) and five females (20%), ranging in age from 31

Table 1
Summary of cases.

Case #	Age/Sex	Clinical history	Drugs	Circumstances	Macroscopic findings	Microscopic findings	Acetone mmol/L	Vitreous glucose mmol/L	Vitreous ketones mmol/L
1	39/F	Type I DM, DKA, Drug abuse	Insulin	Found dead	Cachectic	Hepatic steatosis, Armanni-Ebstein lesion	12.4	–	–
2	31/M	Type I DM	Insulin	Found dead	Fatty liver	Hepatic steatosis, Armanni-Ebstein lesion	8.5	32	8
3	32/M	Drug abuse	Not known	Found dead	Obese, Fatty liver	Hepatic steatosis, Armanni-Ebstein lesion	5.0	48.4	1.5
4	61/F	DM	Insulin	Found dead	None	Hepatic steatosis, Armanni-Ebstein lesion	5.9	–	–
5	37/M	Schizophrenia	Quetiapine	Found dead	Obese, Fatty liver	Hepatic steatosis, Armanni-Ebstein lesion	7.1	36.6	1.5
6	35/M	Type I DM, Schizophrenia	Olanzapine	Found dead	Obese, Fatty liver	Hepatic steatosis, Armanni-Ebstein lesion	7.8	63.7	1.5
7	45/M	Mental illness	Insulin	Found dead	Putrefaction	Hepatic steatosis, Armanni-Ebstein lesion	1.0	–	–
8	58/M	Depression, Obsessivecompulsive disorder	Olanzapine Anti-hypertensives	Found dead	Fatty liver	Hepatic steatosis	1.2	2.5	0.5
9	49/M	DM	Insulin	Found dead	Cirrhosis	Hepatic steatosis Cirrhosis, Armanni-Ebstein lesion, Diabetic nephropathy	7.4	–	–
10	49/M	None	Not known	Found dead	Cachectic, Fatty liver	Hepatic steatosis, Armanni-Ebstein lesion	8.1	12.4	4
11	47/M	Type II DM	Oral-hypoglycemics	Found dead	Fatty liver	Hepatic steatosis	0.5	3.0	N
12	49/M	Type I DM	Insulin	Found dead	Dehydration, Scarred and pitted kidneys	Hepatic steatosis, Armanni-Ebstein lesion, Diabetic nephropathy	7.6	49.8	4
13	40/M	DM, Bipolar disorders	Oral-hypoglycemics, Quetiapine	Found dead	Obese	Hepatic steatosis	3.3	29.3	N
14	42/F	DM	Insulin	Found dead	None	Hepatic steatosis, Armanni-Ebstein lesion	8.5	28.0	4
15	62/M	Type II DM, Hypertension	Anti-hypertensives	Found VSA	Cardiomegaly	Hepatic steatosis, Armanni-Ebstein lesion, Diabetic nephropathy	1.4	–	–
16	49/F	Type I DM, DKA	Insulin	Found dead	Mild liver fibrosis	Hepatic steatosis, Armanni-Ebstein lesion	5.9	–	–
17	38/M	Type I DM	Insulin	Found dead	None	Hepatic steatosis, Armanni-Ebstein lesion	10.5	61.8	–
18	52/M	Type II DM	Not known	Found dead	Renal scarring	Hepatic steatosis, Armanni-Ebstein lesion	6.2	–	–
19	32/M	Type I DM	Insulin	Found dead	Coronary artery disease	Hepatic steatosis, Armanni-Ebstein lesion, Diabetic nephropathy	9.7	–	–
20	48/M	Type I DM	Insulin	Found dead	None	Hepatic steatosis, Armanni-Ebstein lesion	9.5	41.2	4
21	49/F	DM, Schzophrenia	Insulin, Clozapine	Found dead	None	Armanni-Ebstein lesion	12.8	38.4	8
22	43/M	DM, Hypertension	Oral-hypoglycemics, Anti-hypertensives	Found dead	Obese, Fatty liver	Hepatic steatosis, Armanni-Ebstein lesion, Diabetic nephropathy	7.9	41.4	–
23	37/M	Schizophrenia	Olanzapine, Temazepam	Found dead	Dehydration, Fatty liver	Hepatic steatosis, Armanni-Ebstein lesion	4.0	51.8	1.5
24	56/M	Type I DM, alcohol abuse	Oralhypoglycemics	Found dead	Cachectic, Fatty liver, Granular kidneys, Atrophic pancreas	Steatohepatitis Diabetic nephropathy	1.0	2.1	N
25	35/M	None	Not known	Found dead	None	Hepatic steatosis, Armanni-Ebstein lesion	5.2	45.4	1.5

M = Male, F = Female, DM = Diabetes mellitus, DKA = Diabetic ketoacidosis, N = Negative, VSA = Vital signs absent.

years to 62 years, with a mean age of 45 years. Six individuals (24%) were on second generation anti-psychotic drugs (clozapine, olanzapine, and quetiapine) and three of this subgroup (12%) had no history significant of DM.

Autopsies in all 25 cases revealed no anatomical causes of death, but a variety of pathological features were present such as cachexia, obesity, hepatic steatosis, hepatic fibrosis, cirrhosis, granular and contracted kidneys, and atrophic pancreas. Twenty-four cases (96%) showed microscopic features of hepatic steatosis (Fig. 1) and twenty-one (84%) showed the Armanni-Ebstein lesion (Fig. 2) in the kidney. Diabetic glomerulosclerosis was noted only in six individuals (24%). Both hepatic steatosis and the Armanni-Ebstein lesion were seen in twenty cases (80%) while the combination of hepatic steatosis, Armanni-Ebstein lesion and diabetic glomerulosclerosis were noted only in five cases (20%).

Toxicological analysis of blood in all cases revealed acetone concentrations ranging from 0.5 to 12.8 mmol/L with a mean level of 6.4 mmol/L. In seventeen cases (68%), vitreous glucose results ranged from 2.1 to 63.7 mmol/L with a mean level of 35 mmol/L. In fifteen cases (60%), vitreous ketone results varied from negative levels to 8 mmol/L with a mean level of 2.7 mmol/L.

4. Discussion

DKA most frequently occurs in newly diagnosed or previously unknown diabetic individuals and in diabetics who have an underlying concomitant infection or have missed their insulin treatment. The lack of insulin is accompanied by an increase in counter-regulatory hormones such as glucagon, cortisol, epinephrine and growth hormone. As a result, gluconeogenesis and glycogenolysis are enhanced, leading to severe hyperglycemia. Hyperglycemia in turn induces osmotic diuresis, which leads to dehydration and electrolyte imbalance.

In addition, insulin deficiency enhances lypolysis, which releases free fatty acids from adipose tissue into the serum. These free fatty acids are used as an alternative energy source in hepatic mitochondria and form ketone bodies (acetone, beta hydroxybutyrate and acetoacetate) as end-products. The rate at which ketone bodies are formed may exceed the rate at which they are used by peripheral tissues. Thus, progressive accumulation of ketone bodies in blood causes ketonaemia; excess amounts overflow into the urine causing ketonuria. The low blood pH caused by ketonaemia leads to metabolic acidosis (ketoacidosis). Mechanisms of death in DKA include metabolic derangement with profound acidosis (below pH 7), dehydration, electrolyte imbalance and cerebral oedema.

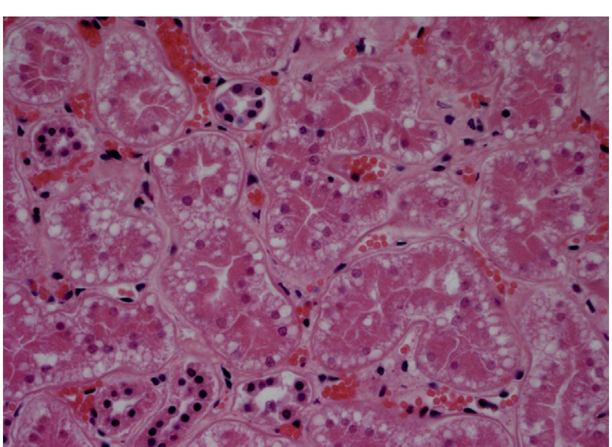

Fig. 1. Armanni-Ebstein lesion (Haematoxylin & Eosin 400×).

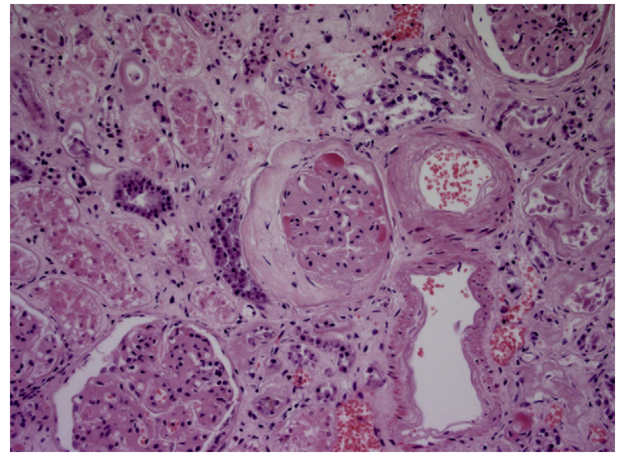

Fig. 2. Diabetic glomerulosclerosis (Haematoxylin & Eosin 200×).

In this study, of the twenty-five cases, Armanni-Ebstein lesions were noted in twenty one (84%). All these cases had high blood acetone levels varying from 1.0 to 12.8 mmol/L. Thirteen of these were tested for vitreous glucose and all had high levels of vitreous glucose ranging from 12.4 to 63.7 mmol/L. In only 4 cases (16%) were no Armanni-Ebstein lesions observed. Of these four, three had relatively low levels of blood acetone ranging from 0.5 to 1.2 mmol/L, and relatively low levels of vitreous glucose ranging from 2.1 to 3 mmol/L. The post-mortem interval in these cases was 1–2 days. These findings imply that Armanni-Ebstein lesions occur when the blood acetone and vitreous glucose levels are relatively high and therefore represent the microscopic counterpart of fatal DKA. This finding conforms to a previous similar study.[10]

Hepatic steatosis occurs in a variety of conditions including DM, protein malnutrition, obesity, anoxia and the presence of toxins. Although hepatic steatosis is a non-specific finding, our observation of the presence of the combination of hepatic steatosis and Armanni-Ebstein lesion in twenty cases (80%) can lead to the determination of DKA as a cause of death in a group of sudden unexpected deaths with otherwise negative autopsies.

A blood acetone level up to 0.35 mmol/L can be normal for a healthy adult.[11] In this study population, the concentration of acetone in the blood exceeded that found in healthy adults. Increased concentrations of acetone can be seen in DKA, ingestion of acetone or isopropanol, a low carbohydrate and high protein diet, starvation, stress or alcohol excess. In our study sample, all individuals were diabetic as suggested either by history or high vitreous glucose levels above 11.1 mmol/L. Coe reviewed over 6000 cases and never found vitreous glucose concentrations >11.1 mmol/L, except in diabetes.[2] Ketoacidosis due to low carbohydrate and high protein diet, starvation, stress and alcohol excess should reveal normal or low glucose levels and be excluded. The histories and investigations did not suggest acetone or isopropanol intoxication.

Post-mortem blood glucose levels are unreliable due to consumption of glucose by bacterial action and tissue glycogenolysis after death.[12] Vitreous glucose is better protected than serum glucose from post-mortem changes and therefore is a good medium for assessment of glucose and ketone bodies, providing more reliable estimates of DKA.[2] In our study sample, seventeen cases (68%) had vitreous glucose results, and fourteen of them had vitreous glucose levels above 11.1 mmol/L. Although three other individuals showed vitreous glucose below 11.1 mmol/L, their relatively high blood acetone levels suggested DKA. Fifteen cases (60%) had vitreous ketone results. In the lab, vitreous ketones were measured by a semi-quantitative analysis using Ketostix® reagent strips. Ketones (as acetoacetic acid) were reported as negative,

0.5 mmol/L, 1.5 mmol/L, 4 mmol/L, 8 mmol/L, or 16 mmol/L. This assay detected acetoacetic acid and acetone but not beta-hydroxybutyric acid. During this method, false positive results could occur in the presence of levadopa or its metaboliltes. Although three of the decedents had negative ketone levels in our study, they had blood acetone levels above the normal limit.

We noted a converse relationship between vitreous ketone concentration and blood acetone concentration. There was no direct relationship between vitreous ketone concentration versus vitreous glucose concentration or blood acetone concentration versus vitreous glucose concentration. Resuscitation significantly increases the glucose concentration in vitreous humour.[13] However, as all the individuals except one in this study were found dead and were not resuscitated, there was no perimortem alteration of glucose levels in their body fluids.

Although second generation anti-psychotic drugs are well known to cause DKA,[14] this concept is not widely recognized by forensic pathologists. This group of drugs can induce DM and DKA by damage to pancreatic islet cells, sympathetic nervous system dysregulation, weight gain and/or increasing insulin resistance.[15–17] In addition, quetiapine may unmask DM.[18] The contribution of these drugs may be easily overlooked.

Except for one (case number 8), the six individuals who were on second generation anti-psychotic drugs demonstrated notably high levels of blood acetone ranging from 3.3 to 12.8 mmol/L and vitreous glucose ranging from 29.3 to 63.7 mmol/L. The highest blood acetone (12.8 mmol/L), vitreous glucose (63.7 mmol/L) and vitreous ketone (8 mmol/L) levels in this case series were demonstrated by this group. This highlights the strong relationship between second generation anti-psychotic drugs and DKA in sudden unexpected deaths with otherwise negative autopsies. Moreover, three of the six did not have a history of DM. This supports the previous suggestion that DKA may be the first presentation in individuals taking second generation anti-psychotic drugs with no past history of DM.[18]

This study recognises and reconfirms that fatal DKA occurs in both type I and II diabetes. The macroscopic autopsy features observed in this study are non-specific and do not guide the pathologist towards the diagnosis of fatal DKA. Once other possibilities have been excluded, the Armanni-Ebstein lesion alone or the combination of hepatic steatosis and Armanni-Ebstein lesion in an otherwise negative autopsy of a sudden unexpected death should raise the suspicion of DKA as the cause of death and indicate biochemical analysis of body fluids. Our findings also remind forensic pathologists to search for fatal DKA in sudden unexpected death with a negative autopsy, where there is a history of second generation anti-psychotic treatment.

Ethical approval

None declared.

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Conflict of interest

No conflict of interest.

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